

RENUMBERED CLAIMS - DRAFT COPY

In the claims:

Please cancel, without prejudice, previously withdrawn claims 4, 5, 18, 19, 24, 25, and 85-91.

Please cancel, without prejudice, claims 6, 20, 21, 22, 33, 64, 70-73, 81-83, and 93.

1. **(Previously presented)** An assay for identifying compounds that mimic a bioactivity of a *hedgehog* protein, comprising:

- (a) providing a cell that transduces intracellular signals of the *hedgehog* pathway and that expresses a naturally occurring *patched* protein;
- (b) contacting the cell with a test compound; and
- (c) detecting activation of the *hedgehog* pathway in the cell by detecting a change in *GLI* expression in the cell, which *GLI* expression is responsive to the *hedgehog* pathway;

wherein a change in the activation of the *hedgehog* pathway in the presence of the test compound, relative to the activation in the absence of the test compound, indicates a *hedgehog*-mimicking activity for the test compound.

2. **(Cancelled)**

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~~3.~~ **(Previously presented)** The assay of claim 1, wherein the cell includes a heterologous nucleic acid recombinantly expressing the *patched* protein.

4-6. **(Cancelled)**

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~~7.~~ **(Previously presented)** The assay of claim ~~3~~, wherein the recombinant cell lacks expression of an endogenous *patched* protein.

8-16. **(Cancelled)**

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~~17.~~ **(Currently amended)** An assay for screening test compounds to identify agents that mimic a bioactivity of a *hedgehog* protein, comprising:

- (a). providing a cell expressing a naturally occurring *patched* protein, which cell transduces intracellular signals of the hedgehog pathway, wherein said *patched* protein binds a naturally occurring *hedgehog* protein;
 - (b). contacting the cell with a test compound; and
 - (c). detecting activation of the *hedgehog* pathway in the cell by detecting a change in GLI expression in the cell, which GLI expression the level of expression of a gene controlled by a GLI transcriptional regulatory element, which GLI transcriptional regulatory element is responsive to the hedgehog pathway,
- wherein a change in the level of activation of the *hedgehog* pathway is indicative of an agent that mimics a bioactivity of a *hedgehog* protein.

18-26. (Cancelled)

5 27. (Previously presented) The assay of claim 17, wherein the cell further comprises a heterologous gene construct encoding the *patched* protein.

28. (Cancelled)

6 29. (Previously presented) The assay of claim 17, wherein the cell further comprises one or more heterologous gene constructs encoding and expressing *costal-2*, *fused* and/or *smoothened* genes.

7 30. (Currently amended) An assay for screening test compounds to identify agents that activate the hedgehog pathway, comprising:

- (a). providing a cell having a recombinant expression vector encoding a naturally occurring mammalian *patched* protein, wherein said cell transduces intracellular signals of the hedgehog pathway;
- (b). contacting the cell with a test compound under conditions wherein the *patched* protein is expressed; and
- (c). detecting a change in the hedgehog pathway in the cell by detecting a change in *GLI* expression in the cell ~~or by detecting a change in the level of expression of a gene~~

controlled by a ~~GLI~~ transcriptional regulatory element, which GLI expression
transcriptional regulatory element is responsive to the hedgehog pathway,
wherein a change in the hedgehog pathway in the presence of the test compound, relative to in
the absence of the test compound, is indicative of an agent that activates the hedgehog
pathway.

31. ~~(Withdrawn)~~ The assay of claim 30, wherein the signal transduction is detected by
detecting a change in phenotype of the cell relative to in the absence of the test compound.

32. (Previously presented) The assay of claim 30, wherein the cell is a human cell.

33. (Cancelled)

34. (Previously presented) The assay of claim 1 or 30, wherein the *patched* protein is of
vertebrate origin.

35. (Previously presented) The assay of claim 34, wherein the *patched* protein is a
mammalian *patched* protein.

36. (Previously presented) The assay of claim 35, wherein the *patched* protein is a human
patched protein.

37-41. (Cancelled)

42. (Previously presented) The assay of any of claims 3, 11, 30, or 38, wherein the cell is a
metazoan cell.

43. (Previously presented) The assay of claim 42, wherein the cell is a mammalian cell.

44. (Previously presented) The assay of claim 42, wherein the cell is an insect cell.

45-46. (Cancelled).

4 7 12 13

21 47. (Currently amended) The assay of claim 1, ~~17~~, 30, or ~~78~~, or 92, wherein the steps of the assay are repeated to screen a library of at least 100 different test compounds.

4 7 12 13

22 48. (Currently amended) The assay of claims 1, ~~17~~, 30, or ~~78~~, or 92, wherein the test compound is selected from small organic molecules or natural product extracts.

4 7 12 13

23 49. (Currently amended) The assay of claim 1, ~~17~~, 30, or ~~78~~, or 92, further comprising preparing a pharmaceutical preparation by combining one or more compounds identified with a biologically acceptable medium.

50-62. (Cancelled)

24 63. (Currently amended) A method for identifying *hedgehog* agonists, comprising: contacting a test agent with cells expressing a naturally occurring *patched* protein, which cells transduce intracellular signals of the hedgehog pathway and wherein said cells undergo a detectable response when contacted with a naturally occurring *hedgehog* protein; and comparing the response of said cells to the test agent with the response of similar cells to a naturally occurring *hedgehog* protein, which detectable response is a change in *GLI* expression in the cell ~~or a change in the level of expression of a gene controlled by a *GLI* transcriptional regulatory element responsive to the hedgehog pathway~~, wherein induction of a response in the presence of the test agent similar to the response induced in the presence of the *hedgehog* protein is indicative of agonist activity of the test agent.

64-65. (Cancelled)

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25 66. (Previously presented) A method of claim ~~63~~, wherein said cells are transfected to express a recombinant form of the *patched* protein.

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26 67. (Previously presented) A method of claim ~~63~~ or 66, wherein said cells are eukaryotic.

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68. (Previously presented) A method of claim 67, wherein said cells are vertebrate cells.

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69. (Previously presented) A method of claim 67, wherein said cells are mammalian cells.

70-77. (Cancelled)

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78. (Currently amended) An assay for screening test compounds to identify agents that antagonize the hedgehog pathway ~~a bioactivity of a hedgehog protein~~, comprising:

- (a). providing a cell expressing a naturally occurring *patched* protein, which cell transduces intracellular signals of the hedgehog pathway and wherein said *patched* protein binds a naturally occurring *hedgehog* protein;
- (b). detecting activation of the hedgehog pathway in said cell by detecting a change in *GLI* expression in the cell ~~or by detecting a change in the level of expression of a gene controlled by a *GLI* transcriptional regulatory element~~, which *GLI expression* ~~transcriptional regulatory element~~ is responsive to the hedgehog pathway;
- (c). contacting the cell with a test compound;
- (d). detecting activation of the *hedgehog* pathway in said cell in the presence of said test compound by detecting a change in *GLI* expression in the cell ~~or by detecting a change in the level of expression of a gene controlled by a *GLI* transcriptional regulatory element~~, which *GLI expression* ~~transcriptional regulatory element~~ is responsive to the hedgehog pathway,

wherein a decrease in the level of activation of the *hedgehog* pathway in the presence of said test compound in comparison to the absence of said test compound is indicative of an agent that antagonizes a bioactivity of a *hedgehog* protein.

79. (Withdrawn) The assay of claim 78, wherein the activation of the *hedgehog* pathway is detected by detecting a change in phenotype of the cell in the presence of the test compound.

80. (Withdrawn) The assay of claim 79, wherein the change in phenotype is detected by detecting gain or loss of expression of a cell-type specific marker.

Cancelled by Examiners' Amendment
Cancelled by Examiners' Amendment

81-91. (Cancelled)

13

~~92.~~ (Currently amended) A method for identifying *hedgehog* antagonists, comprising:
contacting a test compound with cells expressing a naturally occurring *patched* protein, which
cells transduce intracellular signals of the hedgehog pathway and wherein said cells
undergo a detectable response when contacted with a naturally occurring *hedgehog*
protein; and
comparing the response of said cells to the test compound with the response of similar cells to a
naturally occurring *hedgehog* protein; which detectable response is a change in *GLI*
expression in the cell ~~or a change in the level of expression of a gene controlled by a *GLI*~~
~~transcriptional regulatory element responsive to the hedgehog pathway~~, wherein a
decrease in the detectable response in the presence of the test compound in comparison to
the detectable response induced in the presence of the hedgehog protein indicates
antagonist activity of the test compound.

93. (Cancelled)

Please add the following new claims:

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~~94.~~ (New) A method of claim ¹²~~78~~ or ¹³~~92~~, wherein said cells are transfected to express a
recombinant form of the *patched* protein.

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~~95.~~ (New) A method of claim ¹²~~78~~ or ¹³~~92~~, wherein said cells are eukaryotic.

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~~96.~~ (New) A method of claim ¹⁵~~95~~, wherein said cells are vertebrate cells.

17

~~97.~~ (New) A method of claim ¹⁶~~96~~, wherein said cells are mammalian cells.